

LIBRARY
10 JAN 2017

i

**elicitation of natural disease resistance and enhancing the
postharvest quality of *Capsicum annuum* L. through
selected pre-harvest agricultural practices**

A Thesis submitted in partial fulfillment of
the requirements for the degree of

DOCTOR OF PHILOSOPHY

at

Eastern University, Sri Lanka

by

Chandrakantha Mahendranathan



2015

**Faculty of Science,
Eastern University, Sri Lanka,
Chanekalady**

PROCESSED
Maia Library, EUSL



ABSTRACT

Elicitation of natural disease resistance and enhancing the postharvest quality of *Capsicum annuum* L. through selected pre-harvest agricultural practices

A series of studies was undertaken to enhance natural disease resistance of *Capsicum annuum* L. cvs. HYW and CA-8, against anthracnose and postharvest quality, through some selected pre- and postharvest treatments and varying irrigation regimes. *C. acutatum* was shown to be associated with chili anthracnose for the first time in Sri Lanka. The chemical elicitor, potassium silicate (Kasil[®]) was applied as postharvest treatment at a series of concentrations, 0 (control), 100, 200, 400 and 1000 mg/l and found that the concentration at 200 mg/l was effective. Elicitor treatment, at 200 mg/l, was done once in a week for 4 weeks, as pre-harvest soil drench to field grown plants, commencing from flowering. It resulted in significantly reduced anthracnose disease when the harvested fruits were challenge-inoculated with *C. acutatum*. Postharvest application of potassium silicate (Kasil[®]), at the same concentration reduced anthracnose lesion area by 25 - 100%, compared to the untreated controls. Postharvest spray treatment of chili at mature, green stage with potassium silicate at 200mg/l, reduced the severity of anthracnose development by 34-100%. Spore germination assay revealed that potassium silicate has no antifungal effect on conidia of *C. acutatum*. Potassium silicate (Kasil[®]) treated fruits, inoculated with *C. acutatum* after harvest, showed greater accumulation of phytoalexins and enhanced β -1, 3-glucanase activity in the tissues. Phytoalexins and Pathogenesis-Related (PR) Proteins such as β -1, 3-glucanase are considered to play an important role in plant disease resistance.

Deficit irrigation was carried out, where the sweet pepper (*Capsicum annuum* L.) plants cvs. Pepperone, Marconi Rosso and Friggitelto were treated with 100, 200 mL day⁻¹ (lowest and medium irrigations, respectively) and with 400 mL day⁻¹, as control, from the day of anthesis to harvesting maturity. The results revealed that water stress reduced fresh

and dry weights but increased fruit dry matter. Water stress had little effect on the incidence of blossom-end rot (BER). Water stressed plants in fact had a slightly higher incidence of BER than control plants. The concentration of sugars and total phenolics that are linked to taste and/or 'healthfulness' respectively, increased with the less water treatment. Meanwhile, postharvest quality of *C. annuum* L. was enhanced by the influence of soil moisture status, the deficit irrigation.

In conclusion application of chemical elicitor, the Potassium silicate (Kasil[®]), either as a pre-harvest drench or a postharvest spray treatment reduced anthracnose disease in ripe chili showing that Potassium silicate (Kasil[®]) offers a great promise as an alternative control strategy to synthetic pesticides for anthracnose in *C. annuum*. Deficit irrigation improved postharvest quality of *C. annuum*.

TABLE OF CONTENTS

Abstract	iii
Acknowledgements	v
Table of contents	vii
List of Tables	xii
List of Figures	xiv
List of abbreviations	xvi

Chapter – 1

1. Introduction and overview of the research	01
1.1. Research background and objective of the research	03

Chapter – 2

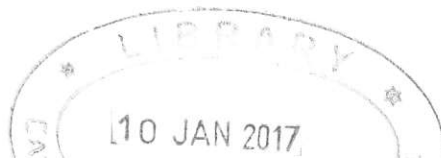
Characterization and pathogenicity of <i>Colletotrichum</i> species associated with <i>Capsicum annuum</i> anthracnose in Sri Lanka	06
2.1 Introduction	06
2.1.1. Anthracnose in <i>Capsicum annuum</i>	06
2.1.2. Causal agents of anthracnose <i>Capsicum annuum</i>	06
2.1.3. Disease cycle and epidemiology of anthracnose caused by <i>Colletotrichum</i> species	10
2.1.4. Management of chili anthracnose	12
2.2. Materials and methods	15
2.2.1. Plant material	15
2.2.2. Collection of diseased samples	15
2.2.3. Isolation of fungi	15
2.2.4. Single spore Isolation	16
2.2.5. Benomyl sensitivity assay for the isolated fungi	16
2.2.6. Growth rate of <i>C.acutatum</i> and <i>C.gleoesporioides</i>	17

2.2.7. Pathogenicity of the organisms isolated from diseased Samples of <i>C. annuum</i>	17
2.2.8. Statistical analysis	18
2.3. Results	19
2.3.2. Collection of diseased Sample	19
2.3.3. Isolation and Identification of fungi from chili anthracnose	20
2.3.4. Single spore isolation	21
2.3.5. Benomyl sensitivity assay for the isolated fungi	22
2.3.6. Growth rate of <i>C. acutatum</i> and <i>C. gleosporioides</i>	22
2.3.7. Pathogenicity of fungi, isolated from diseased chili samples	23
2.4 Discussion	26

Chapter – 3

The effect of Pre- and postharvest treatment of potassium silicate (Kasil®) on anthracnose disease of <i>C. annuum</i>	32
---	-----------

3.1. Introduction	32
3.1.1. Plant defense Mechanisms	32
3.1.2. Induce disease resistance mechanism in plants	32
3.1.2.1. Molecular mechanisms of Induced Resistance	33
3.1.3. Elicitors or plant inducers	34
3.1.3.1. Biotic elicitors or Inducers	35
3.1.3.2. Fungal Elicitors or inducers	36
3.1.3.3. Chemical elicitors or Inducers	40
3.1.4. Effect of induced resistance under field condition	46
3.1.5. Integration and compatibility of plant inducers in crop management	46
3.1.6. Efficacy of inducers under various field conditions	47
3.1.7. Strategies to increase efficacy of Induced Resistance	47



3.2. Materials and methods	49
Effect of Postharvest application of potassium silicate (Kasil[®]) on anthracnose development of <i>C. annuum</i> cvs. HYW and CA-8	49
3.2.1. Potassium silicate (Kasil [®]) treatment and assessment of anthracnose disease development	49
3.2.2. Testing antifungal activity of potassium silicate (Kasil [®])	50
3.2.3. Assessment of induced defense responses in fruit peel due to potassium silicate (Kasil [®]) treatment	51
3.2.3.1. Antifungal compounds	51
3.2.3.2. Defense-related enzymes – β -1, 3-glucanase	52
Effect of Pre-harvest potassium silicate (Kasil[®]) treatment on postharvest anthracnose development in harvested <i>C.annuum</i> Cvs. HYW and CA-8	53
3.2.4. Field preparations and treatment of potassium silicate (Kasil [®])	53
3.2.5. Harvesting	54
3.2.6. Measurement of fruit weight, length and girth.	55
3.2.7. Challenge inoculation and assessment of disease	55
3.3. Results	56
3.3.1. Potassium silicate (Kasil [®]) treatment and anthracnose disease Assessment	56
3.3.2. Testing the antifungal activity of Potassium silicate (Kasil [®])	59
3.3.3. Assessment of induced defense responses in fruits due to potassium silicate (Kasil [®]) treatment	59
3.3.3.1. Antifungal compounds	59
3.3.3.2. Defense- related enzymes - β -1, 3-glucanase	61

Effect of Potassium silicate (Kasil[®]) applied as pre-harvest soil drench on postharvest anthracnose disease development	62
3.3.4. Field preparations and application of potassium silicate (Kasil [®])	62
3.3.5. Harvesting	62
3.3.6. Measurement of fruit weight, length and girth.	63
3.3.7. Challenge inoculation and assessment of disease	63
3.4. Discussion	67

Chapter – 4

Impact of deficit irrigation on the postharvest quality of sweet peppers (<i>Capsicum annuum</i> L.).	74
4.1. Introduction	74
4.2. Material and Methods	77
4.2.1. Plant material and growth conditions	77
4.2.2. Irrigation treatments	77
4.2.3. Soil moisture Content and Environmental monitoring -	78
4.2.4. Fruit sampling	78
4.2.5. Blossom-end rot score	79
4.2.6. Sample Preparation and Reagents.	79
4.2.7. Extraction and Quantification of sugars	79
4.2.8. Extraction and Quantification of Total Phenolics.	80
4.3. Results	81
4.3.1. Soil water status	82
4.3.2. Incidence of blossom-end rots	84
4.3.3. Effect of DI on fruit physiology	86
4.3.4. Effect of deficit irrigation on overall fruit quality	88
4.3.5. Soluble sugars	89
4.3.6. Total Phenols	92